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DOI: https://doi.org/10.1016/j.jgar.2016.11.001

Assessment of animals as a reservoir for colistin resistance: no MCR-1/MCR-2 producing Enterobacteriaceae detected in Swiss livestock

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To the Editor,

The recently described transferable colistin resistance gene *mcr-1* harboured by Enterobacteriaceae poses a threat to the use of polymyxins as a last resort antimicrobial for the treatment of multiresistant bacteria (1). Since its first identification in 2015, numerous retrospective studies have revealed a geographically widespread occurrence of *mcr-1* associated with isolates from humans, animals and retail meat, with its emergence appearing to date back to the 1980s (2). The use of colistin in veterinary medicine and the increasing number of reports on *mcr-1* positive isolates from food-producing animals has focused attention on animals as possible reservoir for the *mcr-1* gene, however, the contribution of livestock to the epidemiology of *mcr-1* is not fully understood (3). Therefore, a study was conducted to evaluate the occurrence of *mcr-1* and *mcr-2* in colistin resistant Enterobacteriaceae isolated from livestock at slaughter in Switzerland. The isolates were collected between April and July 2016 from fecal samples of 325 randomly selected fattening

pigs, 241 randomly selected calves and fecal samples obtained from 100 poultry flocks. Samples of pigs and calves were collected directly after evisceration from the large intestine at slaughterhouse level. Cloacal swabs of chicken were collected at the entry of a poultry slaughterhouse from 10 chicken per flock (approximately 6,000 chicken per flock). For further processing, the 10 swabs were pooled. Swabs were incubated in Enterobacteriaceae enrichment (EE) broth (BD, Franklin Lakes, NJ, USA) at 37°C over night. One loopful of each of the enriched cultures was inoculated onto LB agar plates containing 4mg/l colistin und 5mg/l amphotericin B for selection. Colonies were identified using API ID 32 Etest strips (bioMérieux, Marcyl'Etoile, France) and species with intrinsic resistance to polymyxins (Proteus, Providencia and Morganella) were discarded. Minimal inhibitory concentrations (MICs) of colistin were determined by the broth microdilution according to the guidelines of EUCAST (www.eucast.org). The isolates were evaluated against a panel of further 13 antimicrobial agents using the disk diffusion method. Screening by PCR for *mcr-1* and *mcr-2* was undertaken as described (1, 4).

Overall, 13 (4%) of the fecal swabs from pigs and 8 (3.3%) of the samples from calves yielded non-intrinsic colistin resistant isolates. None of the poultry swabs tested positive. Of the isolates from pigs, 8 (61.5%) were *Hafnia alvei*, three (23%) were *Escherichia coli,* one was *Enterobacter cloacae* and one *Klebsiella pneumoniae* (both 7.7%). Isolates from calves comprised seven (87.5%) *H. alvei* and one (12.5%) *E. coli.* MIC determination confirmed resistance to colistin (MIC>2 mg/L) for all isolates (Table 1). Multidrug resistance (MDR, resistance to three or more classes of antimicrobials) was observed for the *K. pneumoniae* isolate, for three (75%) of the *E. coli* and for five (33.3%) of the *H. alvei* isolates (Table 1). None of the isolates contained the *mcr-1* or *mcr-2* gene. These results indicate that food-producing animals in Switzerland do not represent so far an *mcr-1* or *mcr-2* reservoir. In order to meet the challenges of the rapid ongoing dissemination of plasmid-mediated colistin resistance, a differentiated understanding of its agricultural association is warranted. Future

studies should include the analysis of the role of livestock breeding and food and animal trade on a European as well as global scale.

Funding: This work was supported by the Swiss Federal Office of Public Health

Competing interests: None declared

Ethical approval: Not required

References

1 Liu YY, Wang Y, Walsh TR et al. Emergence of plasmid-mediated colistin resistance mechanism MCR-1 in animals and human beings in China: A microbiological and molecular biological study. Lancet Infect Dis 2016;16:161–168.

2 Schwarz S, Johnson AP. Transferable resistance to colistin: a new but old threat. J Antimicrob Chemother 2016; 71:2066–2070.

3 Poirel L, Nordmann P. Emerging plasmid-encoded colistin resistance: the animal world as the culprit? J Antimicrob Chemother 2016; 71: 2326–2327.

4 Liassine N, Assouvie L, Descombes MC. Very low prevalence of MCR-1/MCR-2 plasmid-mediated colistin resistance from urinary tract Enterobacteriaceae in Switzerland. Int J Infect Dis 2016; 51, 4-5.